Figure 57a – Leu 63→C β chain

```
atgaacgctggtgtcactcagaccccaaaattccaggtcctgaagacaggacaga
gcatgacactgcagtgtgcccaggatatgaaccatgaatacatgtcctggtatcg
acaagacccaggcatggggctgaggctgattcattactcagttggtgctggtatc
actgaccaaggagaagtccccaatggctacaatgtctccagatcaaccacagagg
atttcccgctcaggctgctgtcggctgctcctcccagacatctgtgtacttctg
tgccagcaggccgggactagcgggagggcgaccagagcagtacttcgggccgggc
accaggctcacggtcacagaggacctgaaaaacgtgttcccacccgaggtcgctg
tgtttgagccatcagaagcagagatctcccacacccaaaaggccacactggtgtg
cctggccacaggcttctaccccgaccacgtggagctgagctggtgggtgaatggg
aaggaggtgcacagtggggtcagcacagacccgcagccc▩▩aaggagcagcccg
ccctcaatgactccagatacgctctgagcagccgcctgagggtctcggccacctt
ctggcaggaccccgcaaccacttccgctgtcaagtccagttctacgggctctcg
gagaatgacgagtggacccaggatagggccaaacccgtcacccagatcgtcagcg
ccgaggcctggggtagagcagactaa
```

Figure 57b Leu 63→C β chain

```
M
N₁AGVTQTPKF QVLKTGQSMT LQCAQDMNHE YMSWYRQDPG MGLRLIHYSV
GAGITDQGEV PNGYNVSRST TEDFPLRLLS AAPSQTSVYF CASRPGLAGG
RPEQYFGPGT RLTVTEDLKN VFPPEVAVFE PSEAEISHTQ KATLVCLATG
FYPDHVELSW WVNGKEVHSG VSTDPQPCKE QPALNDSRYA LSSRLRVSAT
FWQDPRNHFR CQVQFYGLSE NDEWTQDRAK PVTQIVSAEA WGRAD*
```

Figure 58a – Glu 15→C β chain

```
atgaacgctggtgtcactcagaccccaaaattccaggtcctgaagacaggacaga
gcatgacactgcagtgtgcccaggatatgaaccatgaatacatgtcctggtatcg
acaagacccaggcatggggctgaggctgattcattactcagttggtgctggtatc
actgaccaaggagaagtccccaatggctacaatgtctccagatcaaccacagagg
atttcccgctcaggctgctgtcggctgctcctcccagacatctgtgtacttctg
tgccagcaggccgggactagcgggagggcgaccagagcagtacttcgggccgggc
accaggctcacggtcacagaggacctgaaaaacgtgttcccacccgaggtcgctg
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cctggccacaggcttctaccccgaccacgtggagctgagctggtgggtgaatggg
aaggaggtgcacagtggggtcagcacagacccgcagcccctcaaggagcagcccg
ccctcaatgactccagatacgctctgagcagccgcctgagggtctcggccacctt
ctggcaggaccccgcaaccacttccgctgtcaagtccagttctacgggctctcg
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ccgaggcctggggtagagcagactaa
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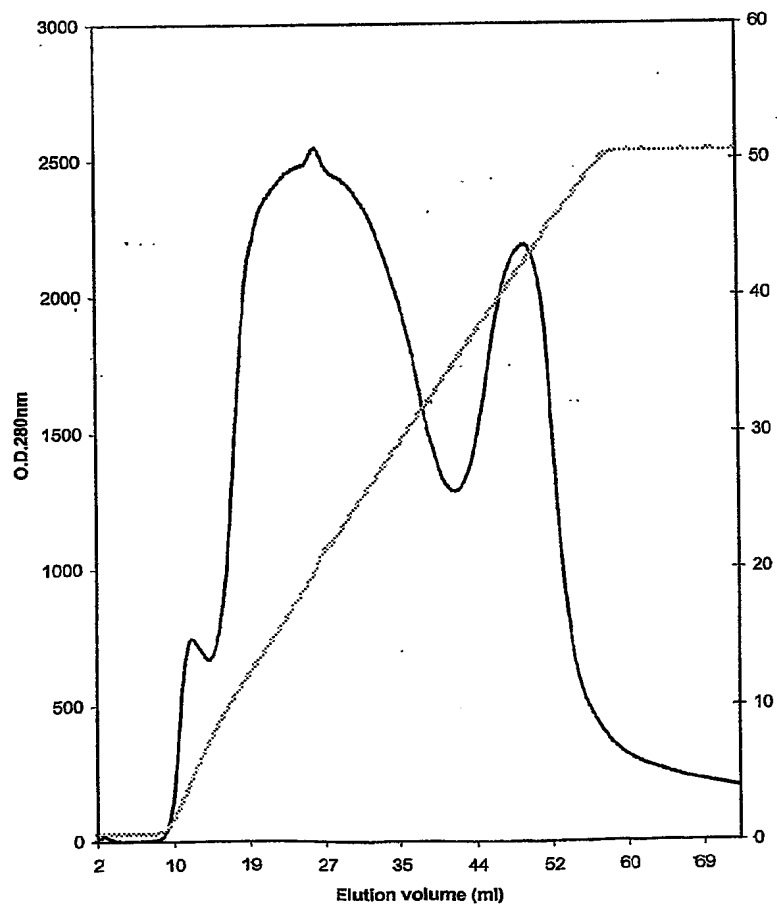
Figure 58b Glu 15→C β chain

```
M
N₁AGVTQTPKF QVLKTGQSMT LQCAQDMNHE YMSWYRQDPG MGLRLIHYSV
GAGITDQGEV PNGYNVSRST TEDFPLRLLS AAPSQTSVYF CASRPGLAGG
RPEQYFGPGT RLTVTEDLKN VFPPEVAVFC PSEAEISHTQ KATLVCLATG
FYPDHVELSW WVNGKEVHSG VSTDPQPLKE QPALNDSRYA LSSRLRVSAT
FWQDPRNHFR CQVQFYGLSE NDEWTQDRAK PVTQIVSAEA WGRAD*
```
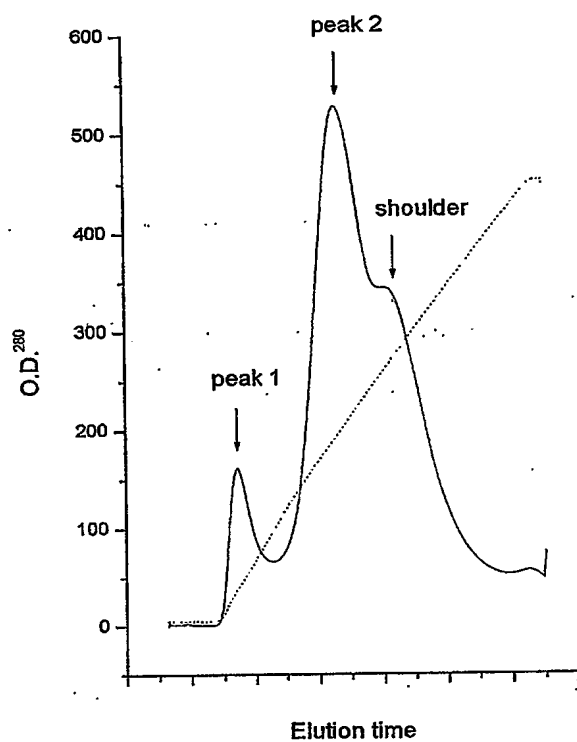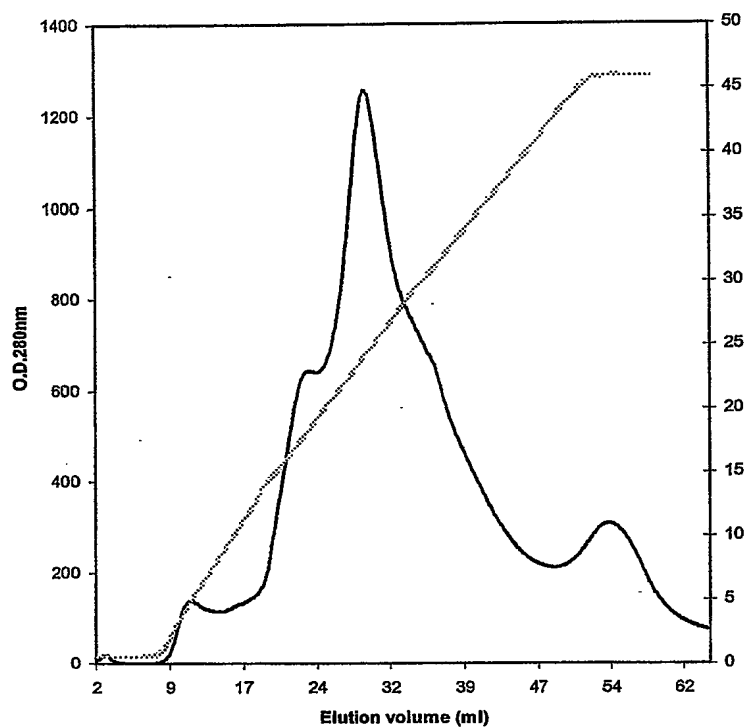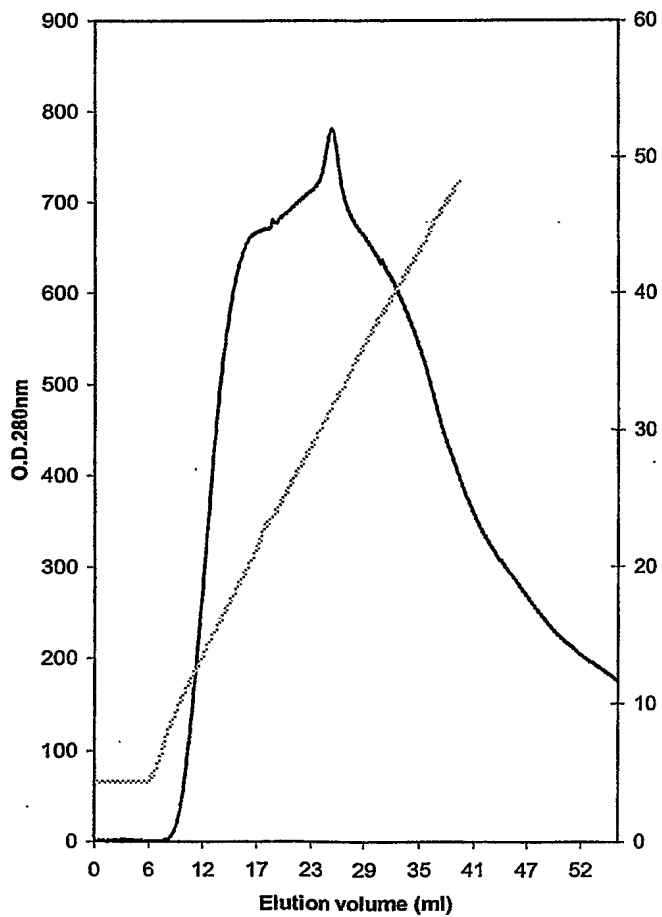
Figure 59: AE column Thr48/Ser57
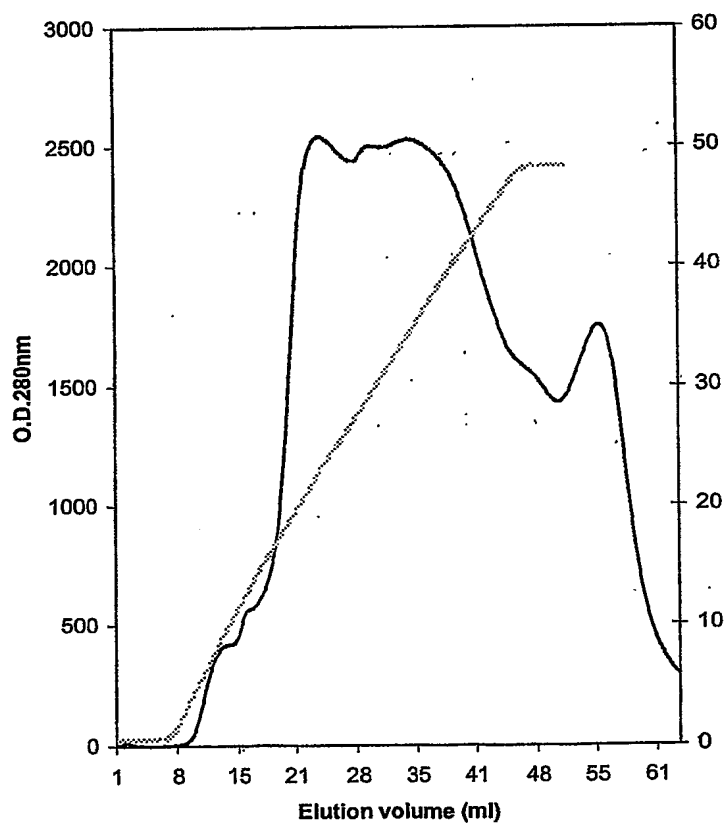
Figure 60: AE column Thr45/Ser77
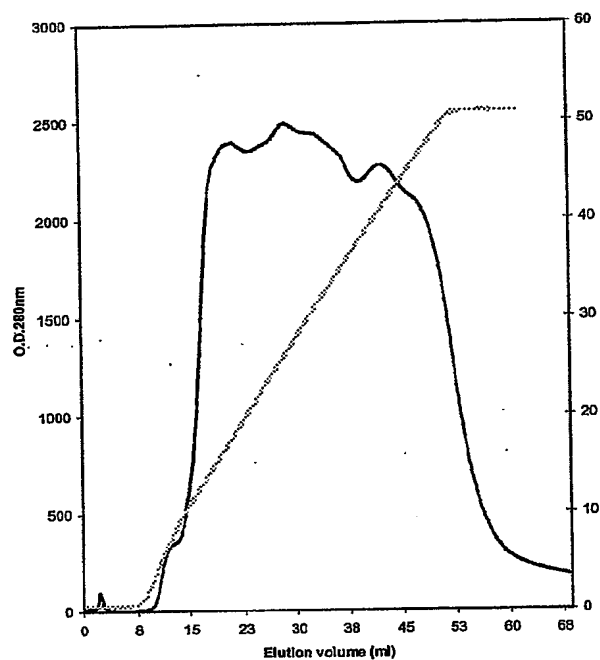
Figure 61: AE column Tyr10/Ser17

Figure 62: AE column Thr45/Asp59

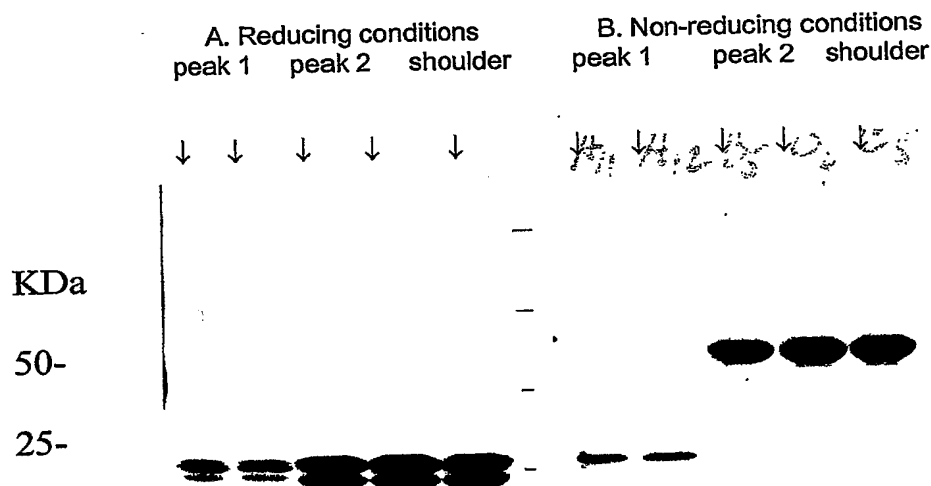Figure 63: AE column Met52/Gly55

Figure 64: AE column Ser15/Glu15

A. Reducing conditions   B. Non-reducing conditions
peak 1   peak 2   shoulder   peak 1   peak 2   shoulder

KDa

50-

25-

Figure 65: SDS PAGE Thr 48 / Ser 57

KDa   A. Reducing conditions   B. Non-reducing conditions

50

25
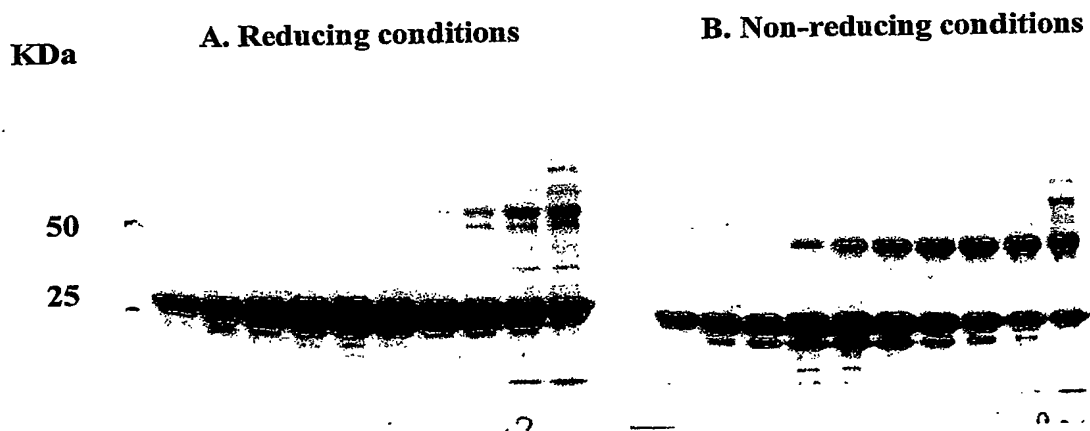
Figure 66: SDS PAGE Thr 45 / Ser 77
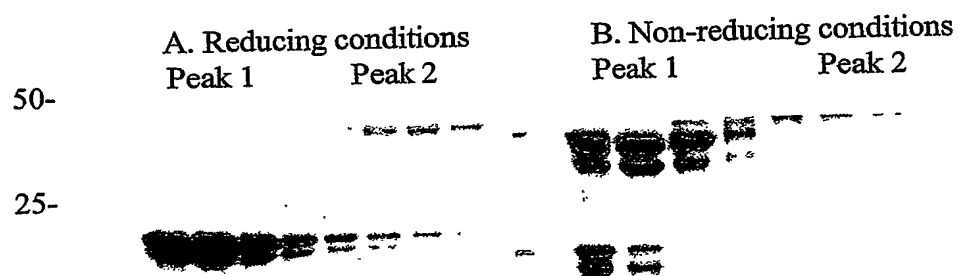
Figure 67 SDS PAGE Tyr 10 / Ser 17
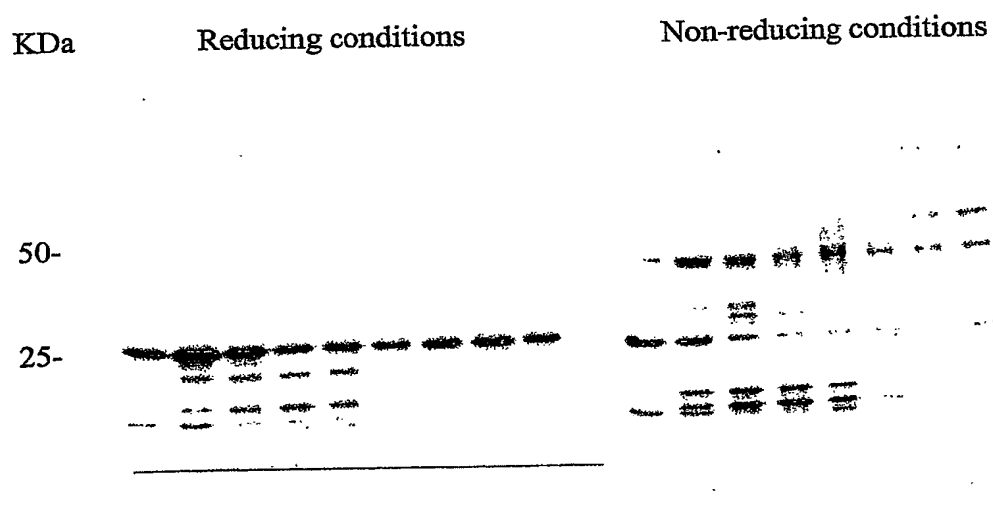
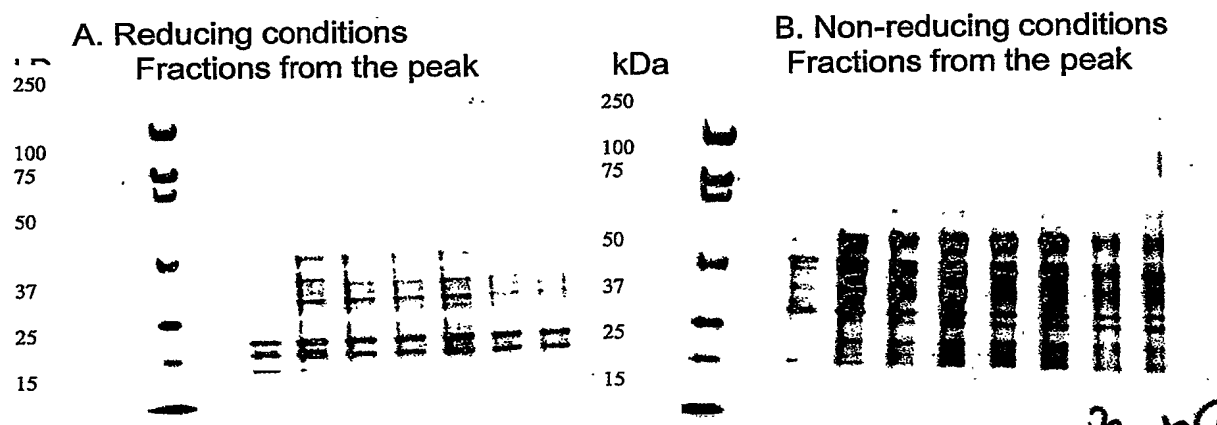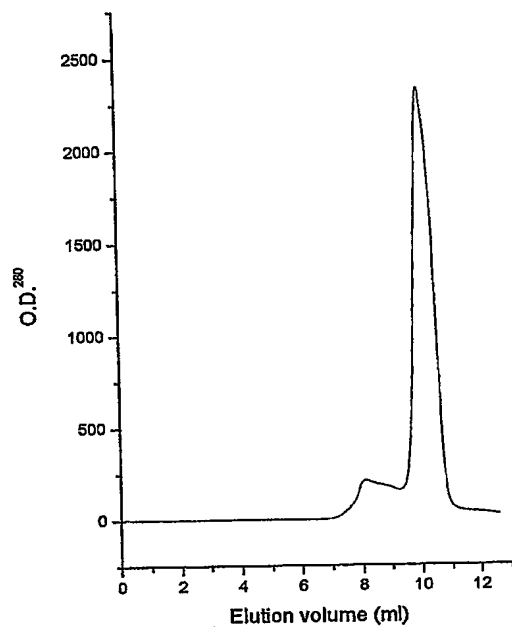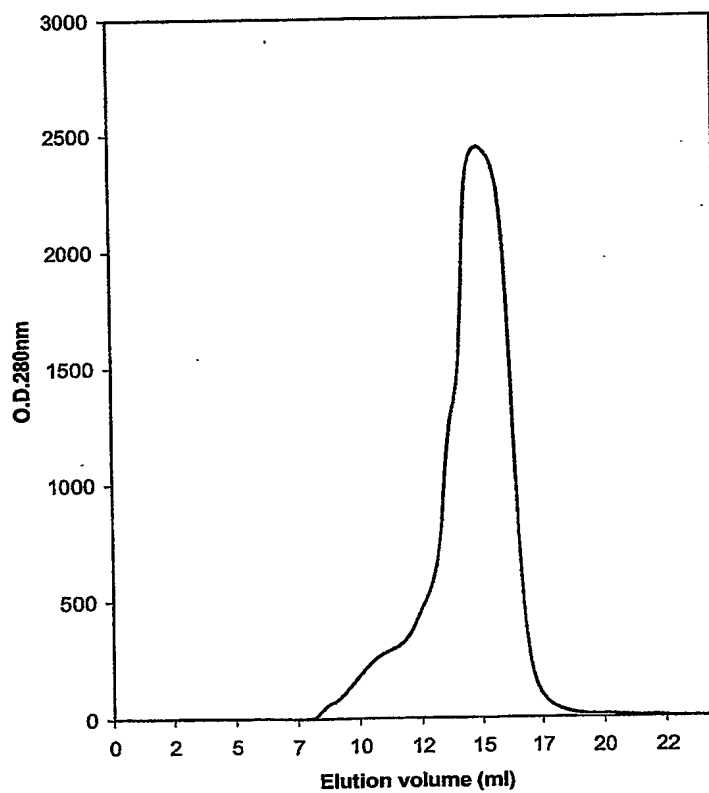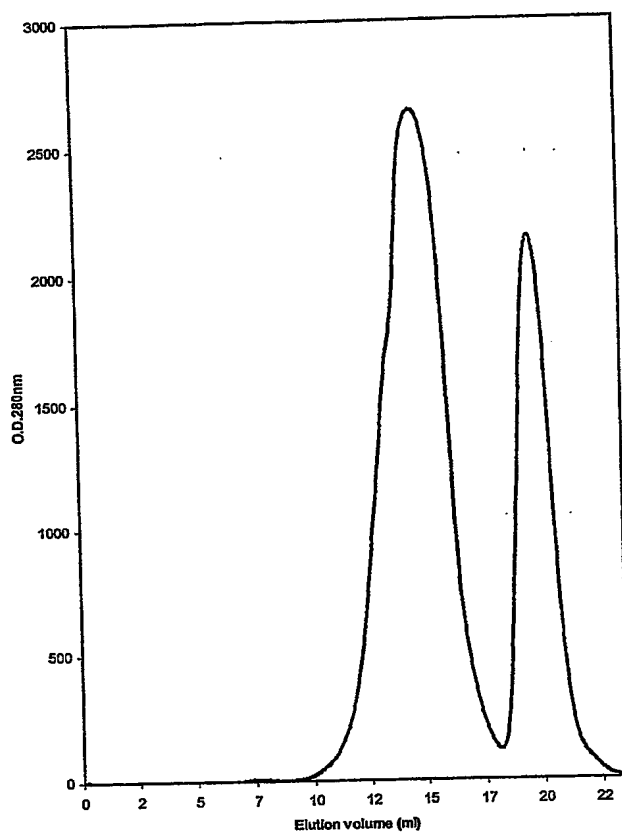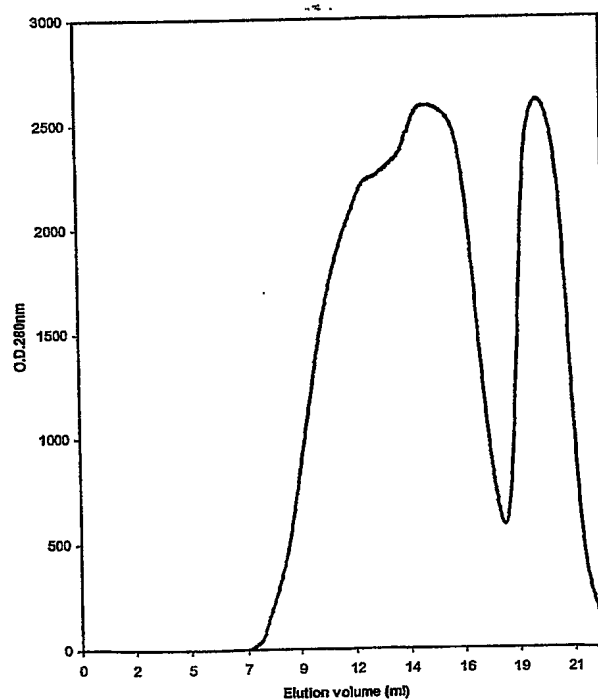Figure 68 SDS PAGE Thr 45 / Asp 59
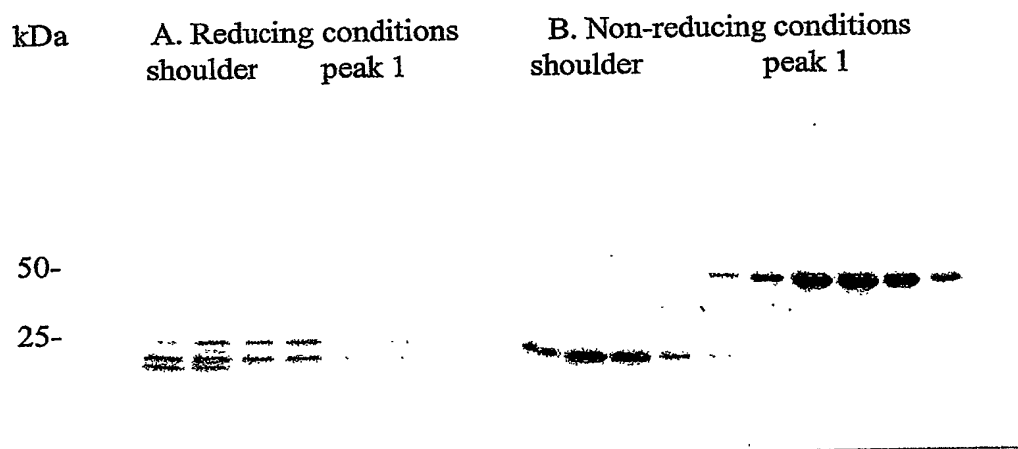
Figure 69: SDS PAGE Met52/Gly55

Figure 70: SDS PAGE Ser 15/Glu 15

Figure 71: SEC Thr48/Ser57

Figure 72: SEC Thr45/Ser77 (200 HR column)

Figure 73: SEC Tyr 10 / Ser 17 (200 HR Column)

Figure 74: SEC Thr 45/Asp 59 (200 HR Column)

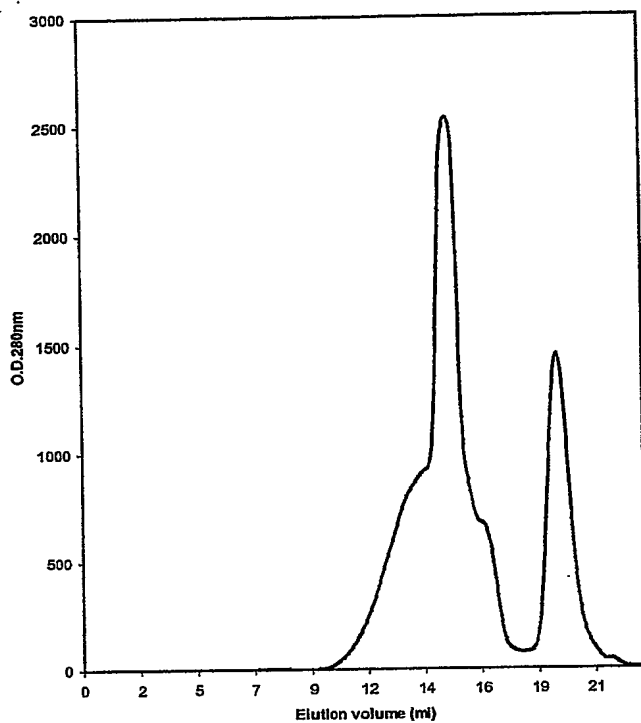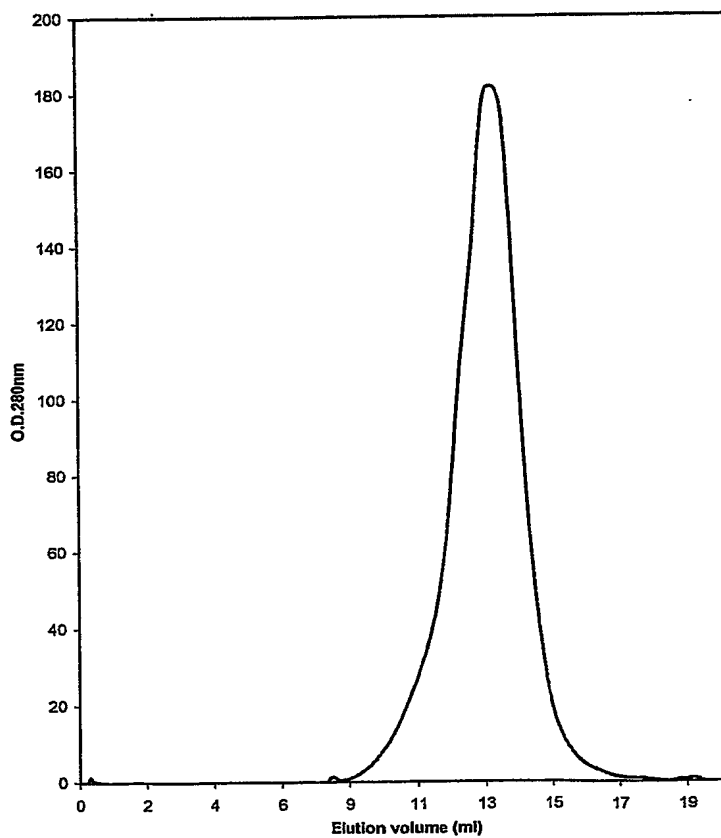**Figure 75 SEC Met 52 / Gly 55 (200 HR Column)**
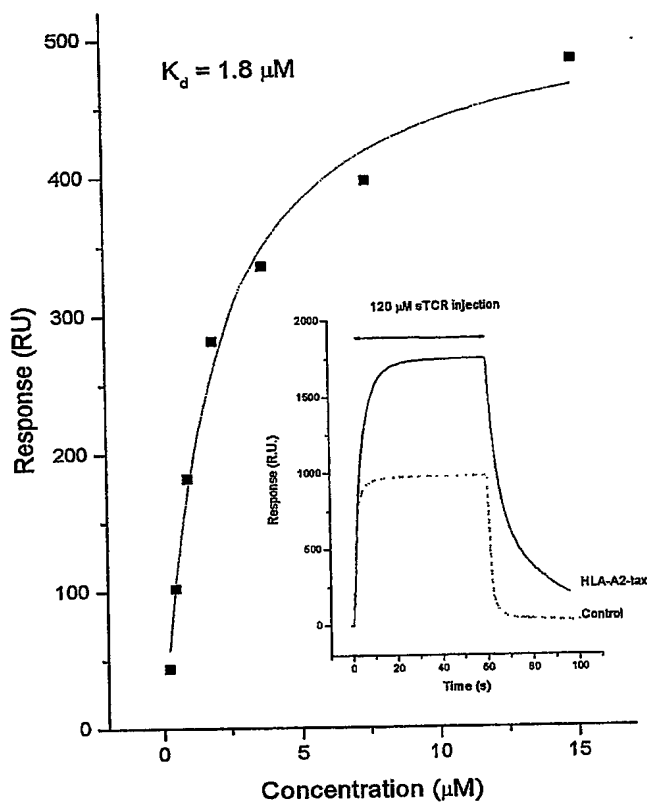

Figure 76: SEC Ser 15/Glu 15 (200 HR Column)
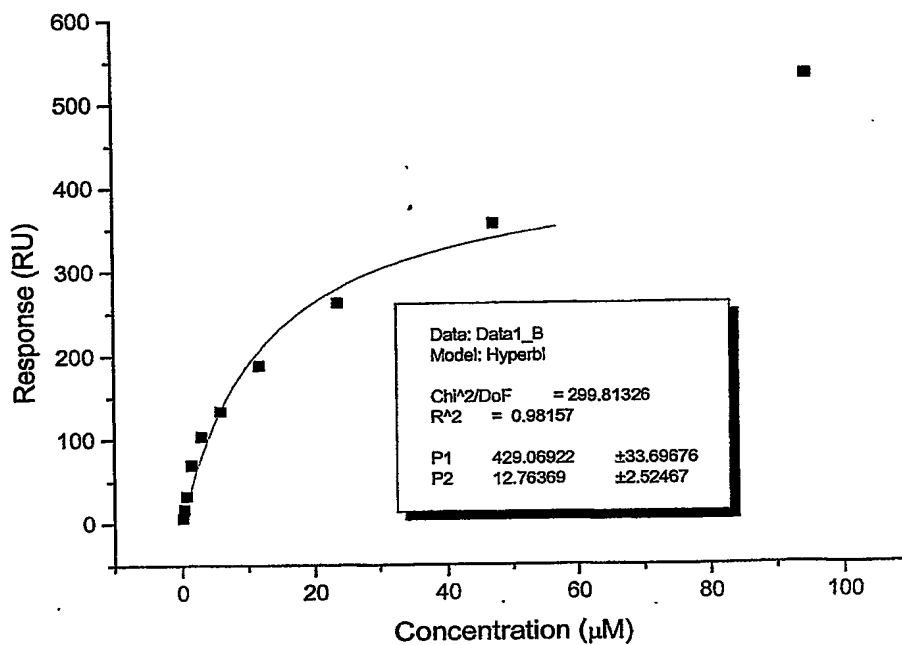
Figure 77: Thr 48/Ser 57

Figure 78: Thr 45/Ser 77
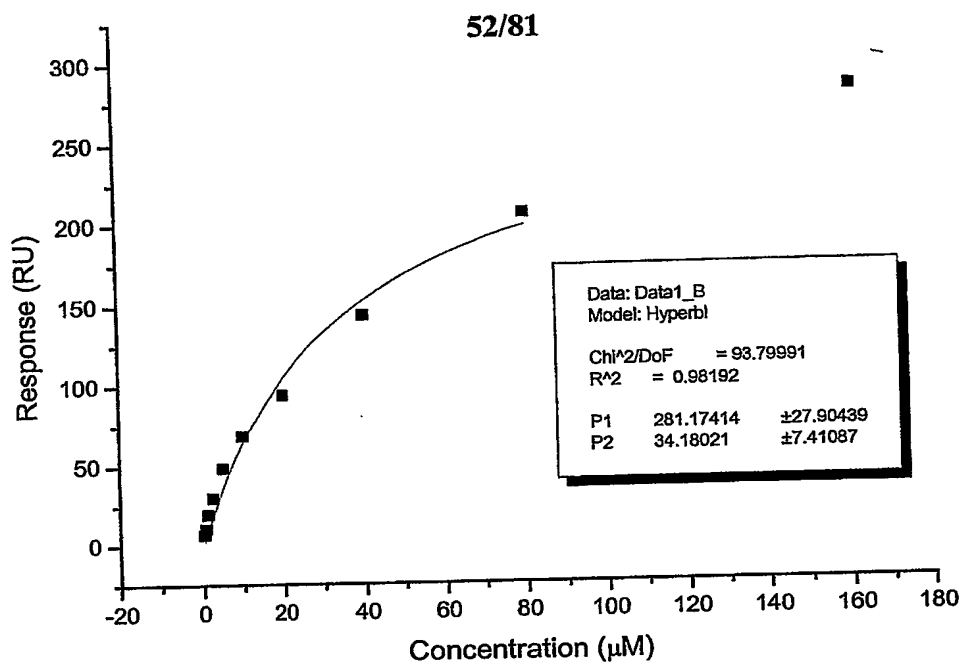
Figure 79: Tyr 10/Ser 17

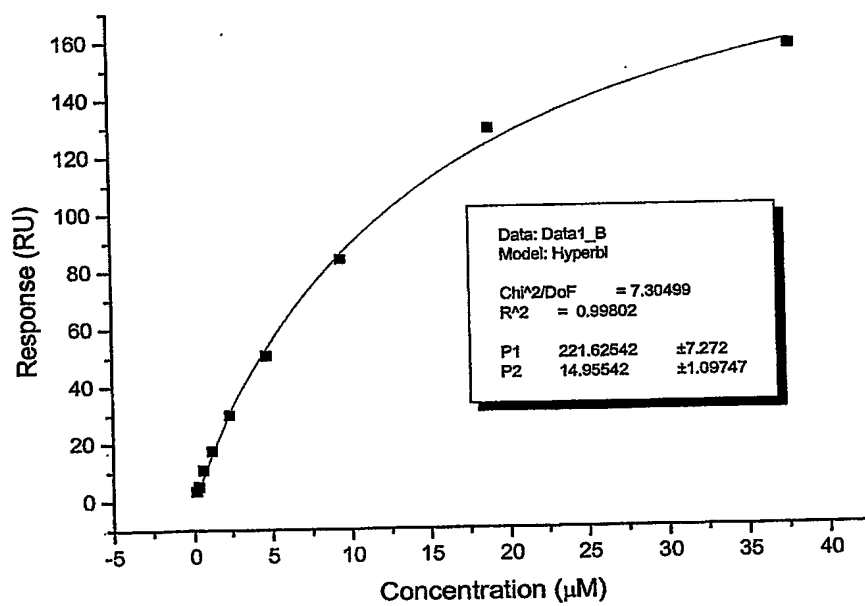**Figure 80: Thr 45/Asp 59**

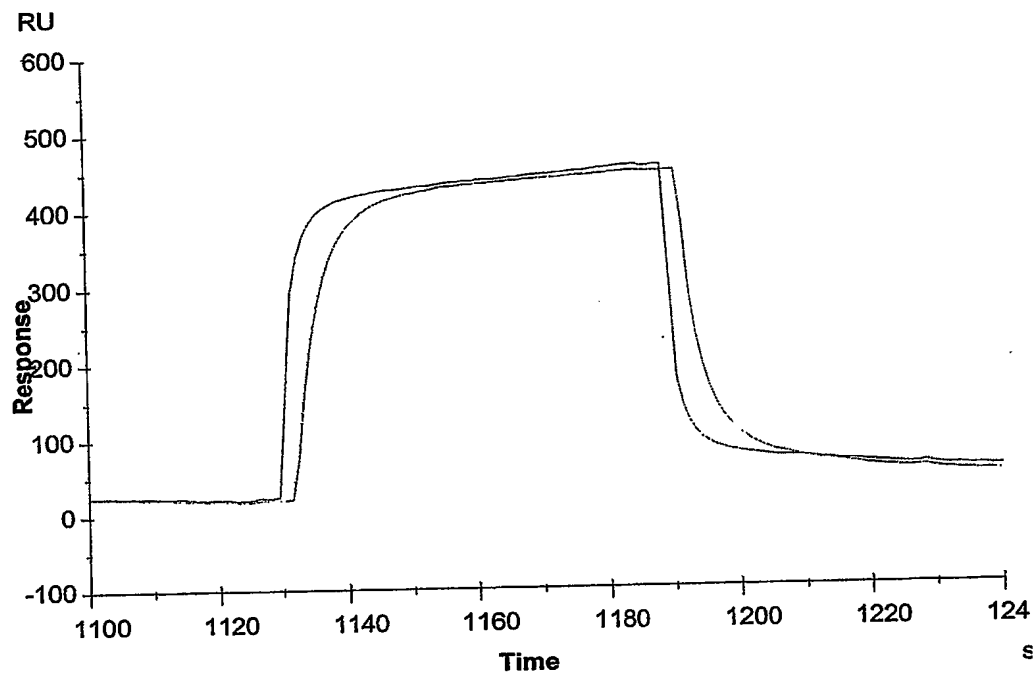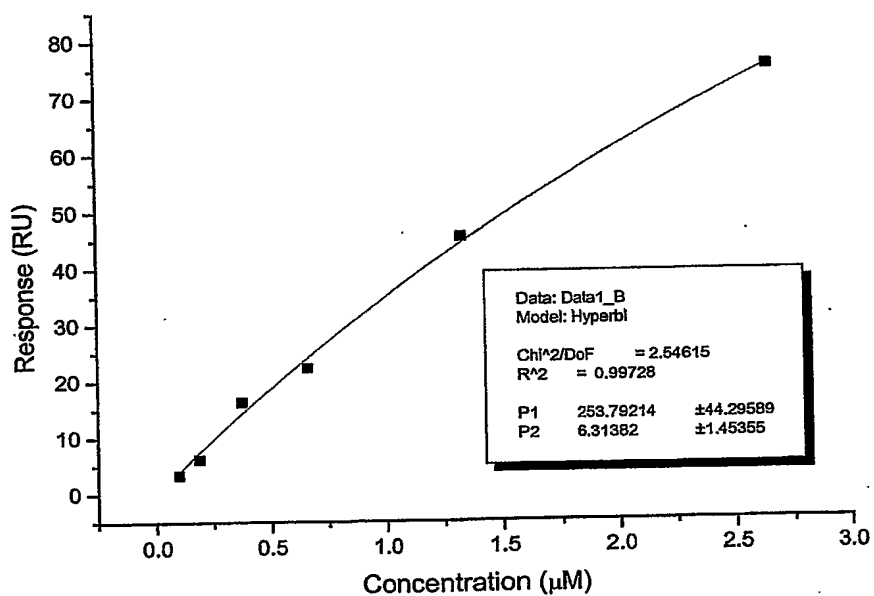**Figure 81: Met 52/Gly 55**

Figure 82: Ser 15/Glu 15

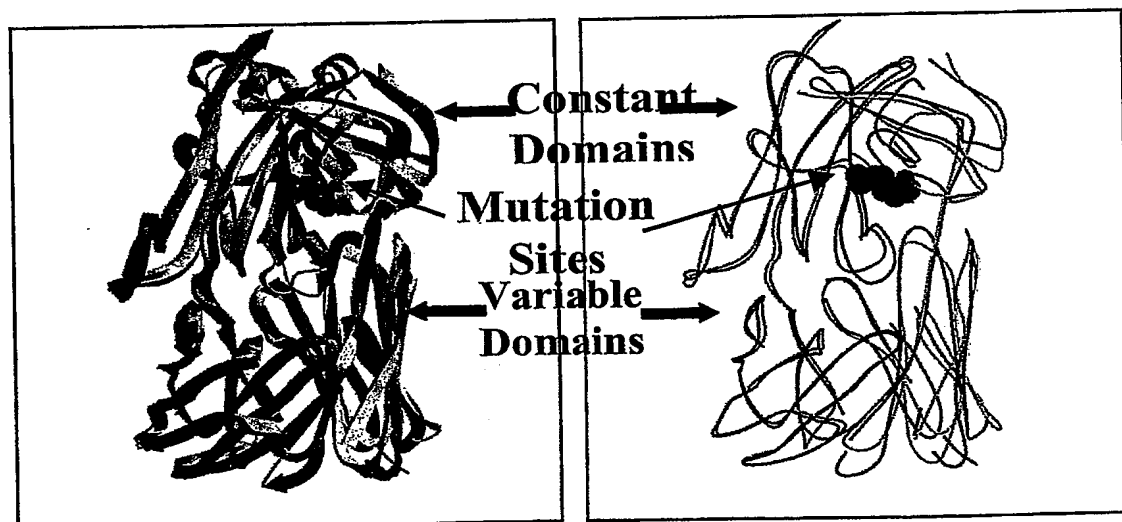Figure 83a

Figure 83b

Figure 84

Figure 85a

atgggtgtcactcagaccccaaaattccaggtcctgaagacaggacagagcatgaca
ctgcagtgtgcccaggatatgaaccatgaatacatgtcctggtatcgacaagaccca
ggcatggggctgaggctgattcattactcagttggtgctggtatcactgaccaagga
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gtccagttctacgggctctcggagaatgacgagtggacccaggatagggccaaaccc
gtcacccagatcgtcagcgccgaggcctggggtagagcagacggatccggtggtggt
ctgaacgatattttgaagctcagaaatcgaatggcattaa

Figure 85b

M G V T Q T P K F Q V L K T G Q S M T L Q C A Q D M N H E Y M
S W Y R Q D P G M G L R L I H Y S V G A G I T D Q G E V P N G
Y N V S R S T T E D F P L R L L S A A P S Q T S V Y F C A S S
Y V G N T G E L F F G E G S R L T V L E D L K N V F P P E V A
V F E P S E A E I S H T Q K A T L V C L A T G F Y P D H V E L
S W W V N G K E V H S G V C T D P Q P L K E Q P A L N D S R Y
A L S S R L R V S A T F W Q D P R N H F R C Q V Q F Y G L S E
N D E W T Q D R A K P V T Q I V S A E A W G R A D G S G G G E
N D I F E A Q K I E W H Stop

Figure 86a

```
atgggtgtcactcagaccccaaaattccaggtcctgaagacaggacagagcatgaca
ctgcagtgtgcccaggatatgaaccatgaatacatgtcctggtatcgacaagaccca
ggcatggggctgaggctgattcattactcagttggtgctggtatcactgaccaagga
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gtcacccagatcgtcagcgccgaggcctggggtagagcagacggatccggtggtggt
catcatcaccatcatcactaa
```

Figure 86b

```
M G V T Q T P K F Q V L K T G Q S M T L Q C A Q D M N H E Y M
S W Y R Q D P G M G L R L I H Y S V G A G I T D Q G E V P N G
Y N V S R S T T E D F P L R L L S A A P S Q T S V Y F C A S S
Y V G N T G E L F F G E G S R L T V L E D L K N V F P P E V A
V F E P S E A E I S H T Q K A T L V C L A T G F Y P D H V E L
S W W V N G K E V H S G V C T D P Q P L K E Q P A L N D S R Y
A L S S R L R V S A T F W Q D P R N H F R C Q V Q F Y G L S E
N D E W T Q D R A K P V T Q I V S A E A W G R A D G S G G G H
H H H H H Stop
```
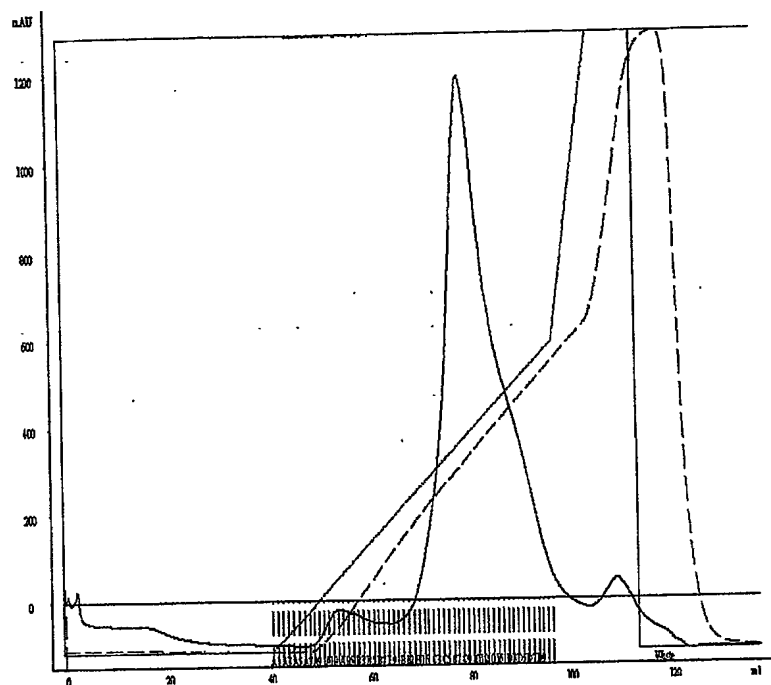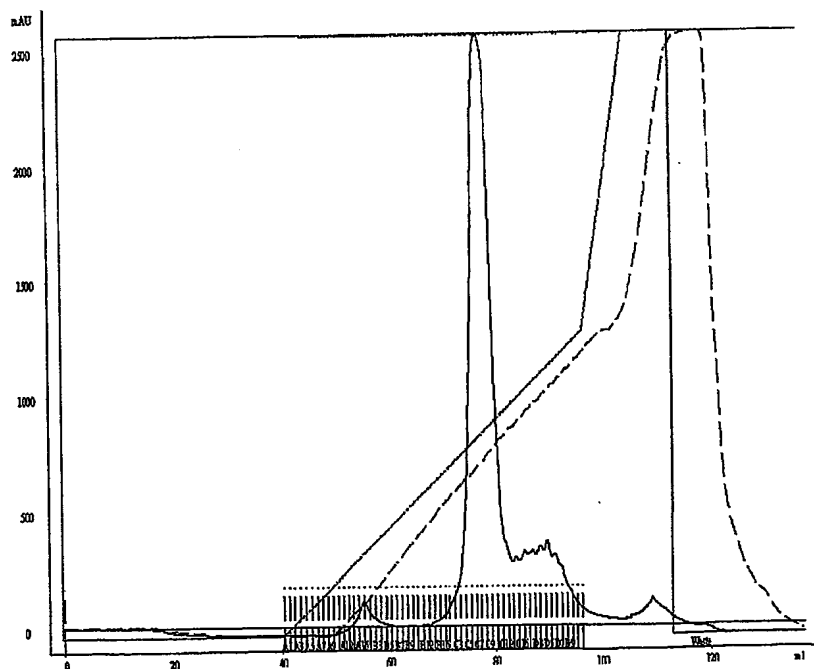
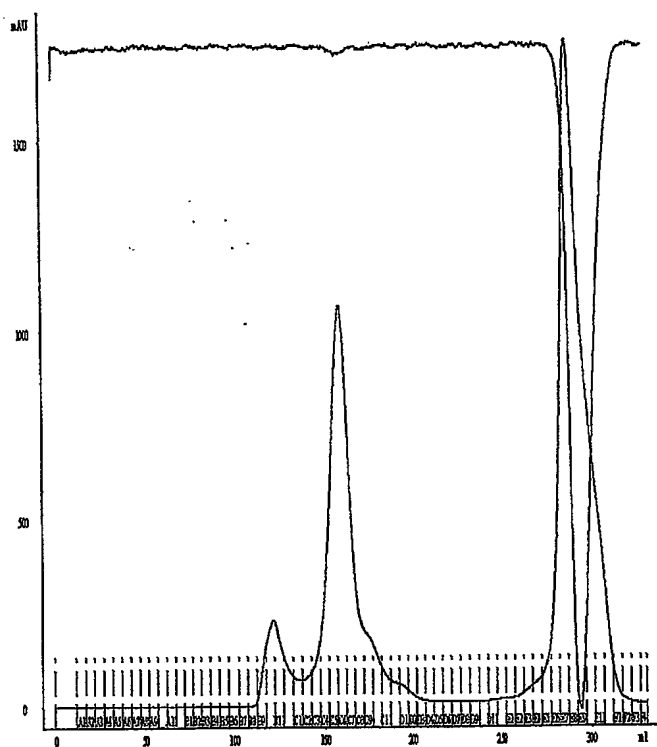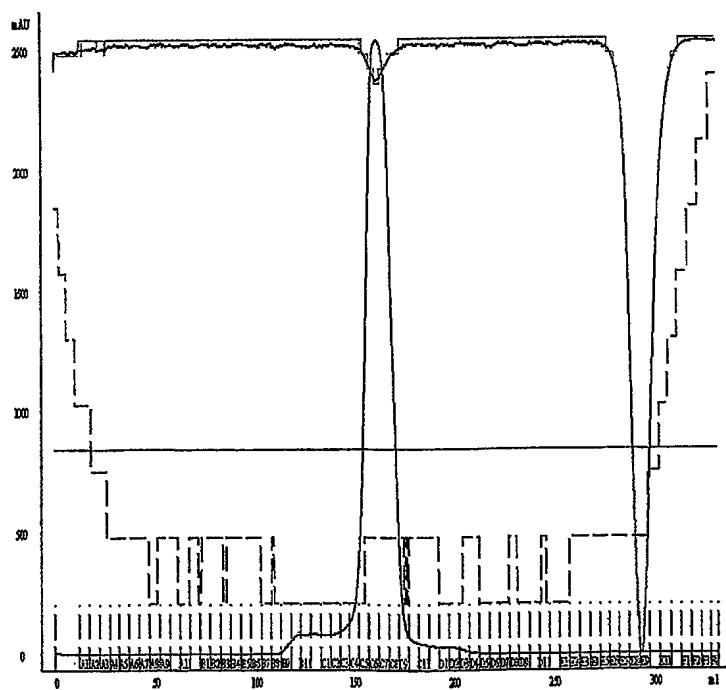## Figure 87

Figure 88

Figure 89
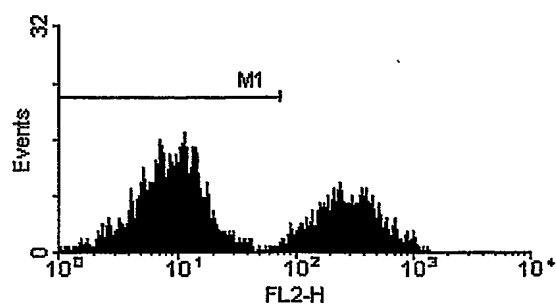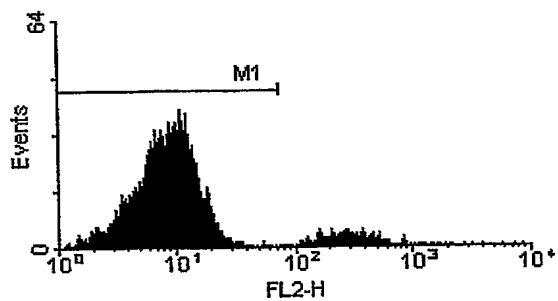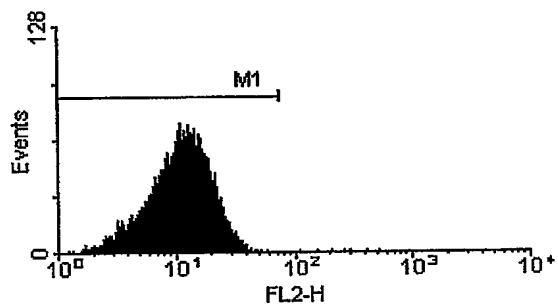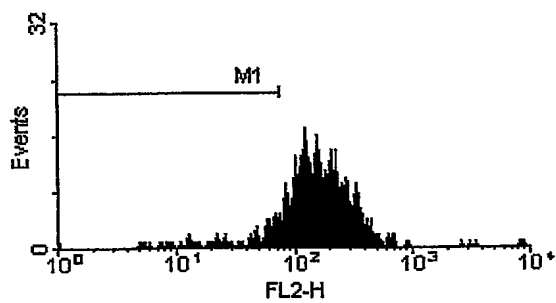

Figure 90
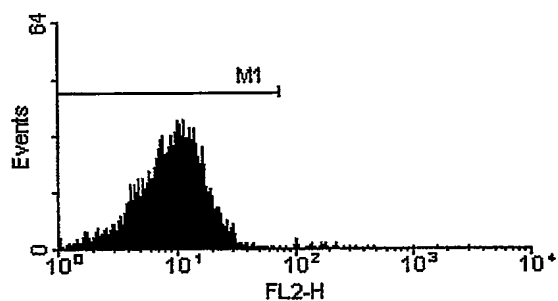
Figure 91a. PP LCL NYESO 0 TCR 5μg
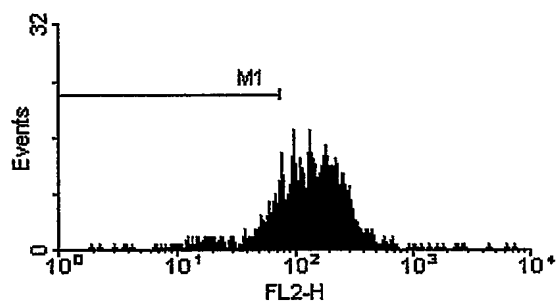
Figure 91b.PP LCL NYESO 10^{-4}M TCR 5μg

Figure 91c.PP LCL NYESO 10^{-5}M TCR 5μg

Figure 91d. PP LCL NYESO 10^{-6}M TCR 5μg

Figure 91e. PP LCL NYESO 0 TCR 10μg

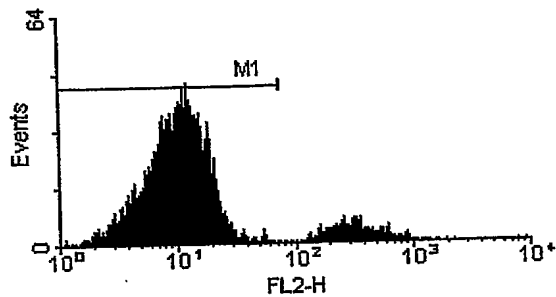**Figure 91f. PP LCL NYESO 10^{-4}M TCR 10μg**

Figure 91g. PP LCL NYESO 10⁻⁵M TCR 10μg

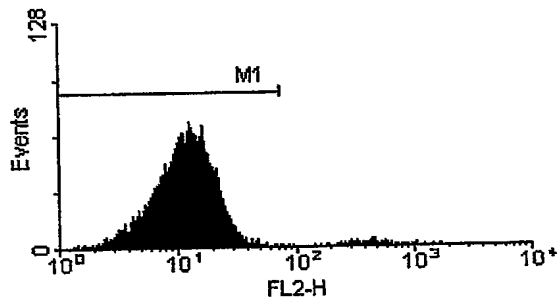**Figure 91h. PP LCL NYESO 10⁻⁶M TCR 10μg**

```
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caaggagaagtccccaatggctacaatgtctccagatcaaccacagaggatttcccg
ctcaggctgctgtcggctgctcctcccagacatctgtgtacttctgtgccagcagg
ccgggactagcgggagggcgaccagagcagtacttcgggccgggcaccaggctcacg
gtcacagaggacctgaacaaggtgttccacccgaggtcgctgtgtttgagccatca
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tctctgagcagccgcctgagggtctcggccaccttctggcaggacccccgcaaccac
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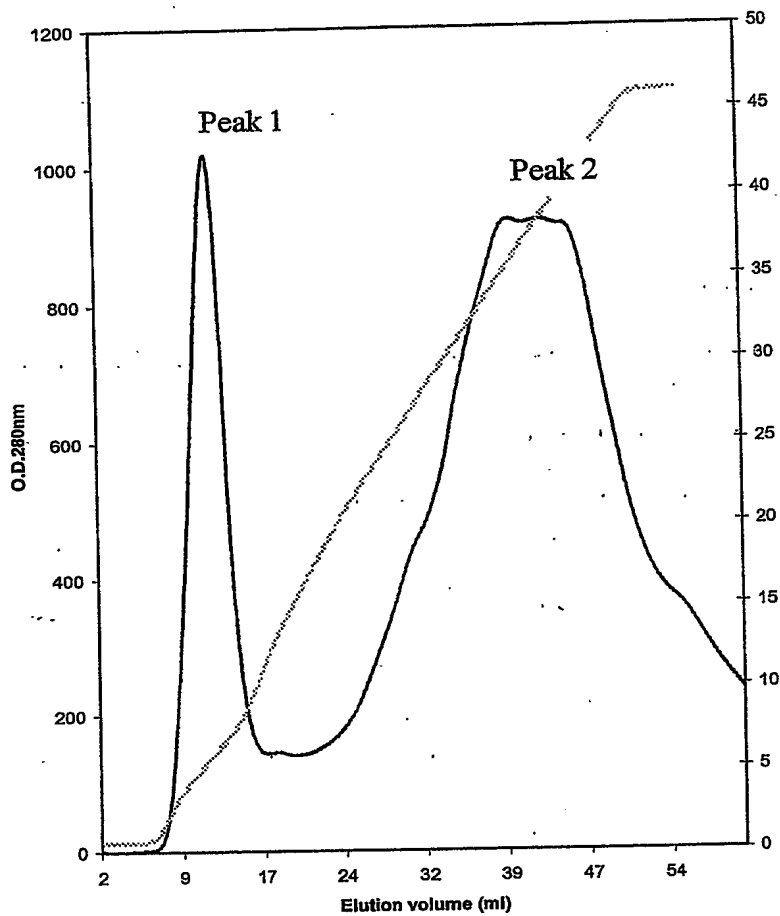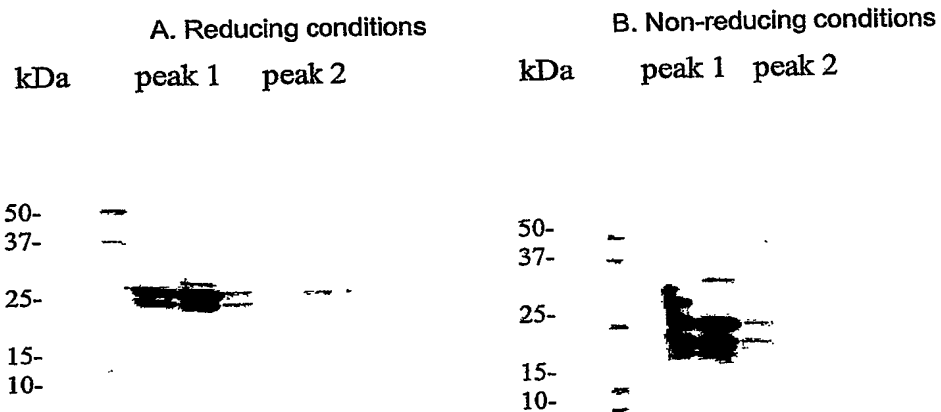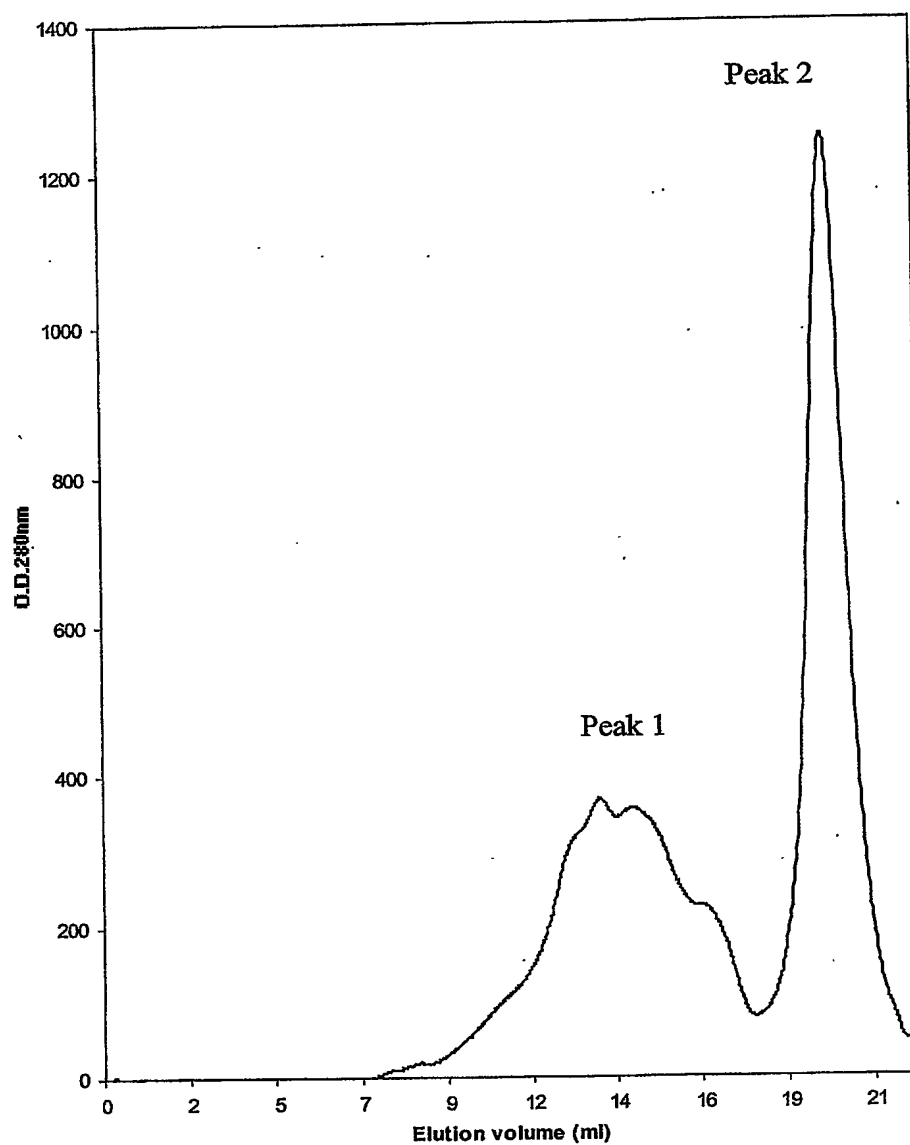
Figure 92

Figure 93

Figure 94

A. Reducing conditions   B. Non-reducing conditions

kDa   peak 1   peak 2   kDa   peak 1   peak 2

Figure 95

Figure 96

A

B